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The views, opinions and/or findings contained in this report are those of the author(s) and should not contrued as an official Department of the Army position, policy or decision, unless so designated by other documentation.

14. ABSTRACT

The overarching goal of our proposal was to build on the success of five Title V programs and initiatives by engaging underrepresented minorities in STEM research using cutting edge equipment. Within the goal, we had two main aims. In Aim one we proposed to use the equipment to enhance five current research programs. In Aim two, we proposed to enhance the quality and capabilities of current courses and allow development of leading edge STEM curricula. In this report, we outline how each piece of requested equipment has been used in research,

15. SUBJECT TERMS

teaching, research, outreach, minority-serving institution, equipment

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Final Report: Shaping the Future: Integrating Cutting Edge Equipment to Enhance Research, Education and Outreach at Nova Southeastern University.

ABSTRACT

The overarching goal of our proposal was to build on the success of five Title V programs and initiatives by engaging underrepresented minorities in STEM research using cutting edge equipment. Within the goal, we had two main aims. In Aim one we proposed to use the equipment to enhance five current research programs. In Aim two, we proposed to enhance the quality and capabilities of current courses and allow development of leading edge STEM curricula. In this report, we outline how each piece of requested equipment has been used in research, teaching and outreach. Specifically, in research, all five faculty, as well as additional faculty, on this proposal have made significant progress on their projects, as evidenced through publications and grant proposals. In teaching, we have successfully integrated the equipment into several classes and have reached over 500 students. We have also participated in outreach with additional minority serving institutions. Overall, we believe that we have exceeded the goals outlined in our original project narrative.

Enter List of papers submitted or published that acknowledge ARO support from the start of the project to the date of this printing. List the papers, including journal references, in the following categories:

(a) Papers published in peer-reviewed journals (N/A for none)

Received   Paper

   TOTAL:

Number of Papers published in peer-reviewed journals:

(b) Papers published in non-peer-reviewed journals (N/A for none)

Received   Paper

   TOTAL:

Number of Papers published in non peer-reviewed journals:

(c) Presentations

American Junior Academy of Science and AAAS meeting (2015) Brigitte Blanco, Puja Patel, Sujay Kamisetty, Robert C. Speth, & James R. Munoz. Neural Stem Cell Proliferation, Differentiation, and Survival in Response to AT1 and AT2 Angiotensin II Receptor-Specific Agonism.

Miami Dade STEMversations Series. 2015. Munoz, James. Current Topics in Neural Stem Cell Research, 2015.

FIU McNair Scholars Research Conference (2015). Zara Khan & James R. Munoz, Examining Migration of Neural Stem Cells in the Adult Mayan Cichlid Brain.

Life Sciences South Florida STEM Research Symposium , Indian River State College (April 2015). Brigitte Blanco, Puja Patel, Sujay Kamisetty, Robert C. Speth, & James R. Munoz, Neural Stem Cell Proliferation, Differentiation, and Survival in Response to AT1 and AT2 Angiotensin II Receptor-Specific Agonism.

Life Sciences South Florida STEM Research Symposium, Indian River State College (April 2015). Thomas, S. Fins, A.I., Craddock, T.J., Tartar, J.L. Chronic Sleep Restriction increases biomarkers of inflammation and stress.

SYNAPSE Meeting of the Palm Beach Chapter of Society for Neuroscience, Max Planck Institute (January 2015). Brigitte Blanco, Puja Patel, Sujay Kamisetty, Robert C. Speth, & Munoz, J.R., "Neural Stem Cell Proliferation, Differentiation, and Survival in Response to AT1 and AT2 Angiotensin II Receptor-Specific Agonism."

Nova Southeastern University, Ft Lauderdale, FL. (February 2015) Smith, R.P. (2015) Engineering a Synthetic Nematode-Killing Bacteria. Science Colloquium Series.

Life Science South Florida Undergraduate Student Symposium (April 2015) Bracho O, Manchery C, Haskell EC, Blanar C, and Smith RP. Dynamic regulation of toxic engineered bacteria prevents learning in the model nematode Caenorhabditis elegans.

Life Science South Florida Undergraduate Student Symposium (April 2015). McKissack H, Mohammed U, Cahill K, Nemzer L, Smith RP. Investigating the Causes of Antibiotic Resistance in a Periodic Environment.

Nova Southeastern University Undergraduate Student Symposium (April 2015). Pandya D, Smith, RP, Blanar C, Haskell EC. "Modeling and Simulation of Caenorhabditis elegans Chemotaxis in Response to a Dynamic Engineered Bacteria."

Nova Southeastern University Undergraduate Student Symposium (April 2015) McKissack H, Mohammed U, Cahill K, Nemzer L, Smith RP. Investigating the Causes of Antibiotic Resistance in a Periodic Environment. .

Nova Southeastern University Undergraduate Student Symposium (April 2015) Pandya D, Smith, RP, Blanar C, Haskell EC. "Modeling and Simulation of Caenorhabditis elegans Chemotaxis in Response to a Dynamic Engineered Bacteria." .

Nova Southeastern University Undergraduate Student Symposium (April 2015) Bracho O, Manchery C, Hakell EC, Blanar C, and Smith RP. Dynamic regulation of toxic engineered bacteria prevents learning in the model nematode Caenorhabditis elegans. .

Number of Presentations: 13.00

Non Peer-Reviewed Conference Proceeding publications (other than abstracts):

Received   Paper

TOTAL:

Number of Non Peer-Reviewed Conference Proceeding publications (other than abstracts):

Peer-Reviewed Conference Proceeding publications (other than abstracts):

Received   Paper

04/30/2015   1.00   Divya Pandya, Christopher Blanar, Robert P Smith , Evan Haskell. Modeling and simulation of
                    Caenorhabditis elegans chemotaxis in response to a dynamic engineered bacteria,
                    Proceedings of European Council for Modeling and Simulation. 26-MAY-15, . : ,

   TOTAL: **1**

Number of Peer-Reviewed Conference Proceeding publications (other than abstracts):

(d) Manuscripts

Received   Paper

   TOTAL:

Number of Manuscripts:

Books

Received   Book

   TOTAL:

TOTAL:

Patents Submitted

We have not submitted patents.

Patents Awarded

We have not been awarded patents.

Awards

Honorable Mention: Nova Southeastern University's 2015 Undergraduate Symposium.
Brigitte Blanco, Puja Patel, Sujay Kamisetty, Robert C. Speth, & Munoz, J.R., "Neural Stem Cell Proliferation, Differentiation, and Survival in Response to AT1 and AT2 Angiotensin II Receptor-Specific Agonism."

Honorable Mention: Nova Southeastern University's 2015 Undergraduate Symposium.
Bracho O, Manchery C, Hakell EC, Blanar C, and Smith RP. Dynamic regulation of toxic engineered bacteria prevents learning in the model nematode Caenorhabditis elegans. Nova Southeastern University Undergraduate Student Symposium

3rd place poster: Nova Southeastern University's 2015 Undergraduate Symposium.
McKissack H, Mohammed U, Cahill K, Nemzer L, Smith RP. Investigating the Causes of Antibiotic Resistance in a Periodic Environment. Nova Southeastern University Undergraduate Student Symposium

2nd place poster: Life Sciences South Florida STEM Research Symposium , Indian River State College, April 2015.
Pandya D, Smith, RP, Blanar C, Haskell EC. Modeling and Simulation of Caenorhabditis elegans Chemotaxis in Response to a Dynamic Engineered Bacteria." Nova Southeastern University Undergraduate Student Symposium

Graduate Students

NAME	PERCENT_SUPPORTED	Discipline
Cortney Wilson	0.00	
Lauren Hill	0.00	
Shaina Fieldstone	0.00	
Margaret Smith	0.00	
Andrea Lopez	0.00	
Lauren O'Connell	0.00	
Renee Potens	0.00	
Nidhi Vijayan	0.00	
FTE Equivalent:	**0.00**	
Total Number:	**8**	

Names of Post Doctorates

NAME	PERCENT_SUPPORTED
FTE Equivalent:	
Total Number:	

Names of Faculty Supported

NAME	PERCENT_SUPPORTED	National Academy Member
Robert P Smith	0.05	
Jose V Lopez	0.05	
Jaime Tartar	0.05	
Aurelien Tartar	0.05	
James Munoz	0.05	
Robert C Speth	0.00	
Evan Haskell	0.00	
Christopher Blanar	0.00	
Josh Loomis	0.00	
Emily Schmitt	0.00	
Aarti Raja	0.00	
Reza Rezaghifard	0.00	
Dimitri Giarikos	0.00	
Naomi D'Allesio	0.00	
Julie Torruellas Garcia	0.00	
Omar Eldakar	0.00	
Louis Nemzer	0.00	
George Duncan	0.00	
Nwadiuto Esiobu	0.00	
Ana I Fins	0.00	
Travis Craddock	0.00	
Mercedes Fernandez	0.00	
New Entry	0.00	
FTE Equivalent:	**0.25**	
Total Number:	**23**	

Names of Under Graduate students supported

NAME	PERCENT_SUPPORTED	Discipline
Sujay Kamisetty	0.00	Biology
Puja Patel	0.00	Biology
Divya Pandya	0.00	Biology
Cyril Manchery	0.00	Biology
Olena Bracho	0.00	Biology
Uzair Mohammed	0.00	Biology
Haley McKissack	0.00	Biology
Steven Thomas	0.00	Biology
Josue Conde	0.00	Biology
Amal Bhullar	0.00	Biology
Asheley Chang-Story	0.00	Biology
Louben Dorval	0.00	Biology
Yassmen Etayem	0.00	Biology
Rebecaa Quinn	0.00	Biology
Samuel Thomas	0.00	Biology
Mateo Castro	0.00	Biology
Samantha Sandor	0.00	Biology
Neha Siddiqui	0.00	Biology
Franklin Hifferman	0.00	Biology
Sumayyah Abiff	0.00	Biology
Katrina Fins	0.00	Biology
~400 BIOL 1500 (Introductory Biol	0.00	
~80 BIOL 3600 (Genetics) Studen	0.00	
30 BIOL 3400 (Microbiology) Stud	0.00	
20 BIOL 3900 (Parasitology) Stude	0.00	
20 NEUR 2500 (Intro to Neuroscie	0.00	
11 NEUR 3000 (Behavioral Geneti	0.00	
13 NEUR 2700 (Methods in Neuro	0.00	
6 PSYC 5100 (Behavioral Psychol	0.00	
FTE Equivalent:	**0.00**	
Total Number:	**29**	

Student Metrics
This section only applies to graduating undergraduates supported by this agreement in this reporting period

The number of undergraduates funded by this agreement who graduated during this period: 39.00

The number of undergraduates funded by this agreement who graduated during this period with a degree in science, mathematics, engineering, or technology fields:...... 39.00

The number of undergraduates funded by your agreement who graduated during this period and will continue to pursue a graduate or Ph.D. degree in science, mathematics, engineering, or technology fields:...... 0.00

Number of graduating undergraduates who achieved a 3.5 GPA to 4.0 (4.0 max scale):...... 185.00

Number of graduating undergraduates funded by a DoD funded Center of Excellence grant for Education, Research and Engineering:...... 0.00

The number of undergraduates funded by your agreement who graduated during this period and intend to work for the Department of Defense 0.00

The number of undergraduates funded by your agreement who graduated during this period and will receive scholarships or fellowships for further studies in science, mathematics, engineering or technology fields:...... 0.00

Names of Personnel receiving masters degrees

NAME
Gobin, C.M.
Total Number: 1

Names of personnel receiving PHDs

NAME

Total Number:

Names of other research staff

NAME   PERCENT_SUPPORTED

FTE Equivalent:
Total Number:

Sub Contractors (DD882)

Inventions (DD882)

Scientific Progress

See attachment for scientific progress report.

Technology Transfer

We have no technology transfer to report.

April 30th, 2015

Fred Gregory, Ph.D.
ARO Grants Officer's Representative
100 Alabama Street, NW
Suite 4R15,
Atlanta, GA, 30303-3104

Re: Final Report for Award W911NF-14-1-0070

Dear Dr. Gregory:

We would like to sincerely thank the Army Research Office (ARO) for their support of our proposal entitled "Shaping the Future: Integrating Cutting Edge Equipment to Enhance Research, Education and Outreach at Nova Southeastern University."

Over the past year, our team of researchers has worked diligently to acquire, install and operate each piece of equipment requested. Through the use of cost savings, the amount, and quality, of equipment that we have been able to acquire has exceeded the expectations outlined in our grant proposal. Furthermore, we have used this equipment to accomplish and exceed our research, teaching and outreach goals as described in our original proposal. More importantly however, the integration of this equipment in these three areas has been established in such a way that we envision continuing to use and expand the use of this equipment in the years to come.

In the following report, we have listed what has been accomplished to date using the equipment that has been received. I believe that our accomplishments to date serve as a solid foundation upon which we can continue to build a prolific research and teaching program at NSU. Please note that we have several publications in preparation that have used this equipment. These are listed in Appendix 1. Furthermore, we have prepared and submitted several grant proposals using preliminary data produced using this equipment. These are listed in Appendix 2.

Please feel free to contact me should you require additional information. I look forward to continuing a strong relationship with the Department of Defense and the Army Research Office.

Sincerely,

Robert P Smith, Ph.D.

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List of Figure and Tables

Statement of Problem Studied

The overarching goal of our proposal was to build on the success of five Title V programs and initiatives by engaging underrepresented minorities in STEM research using cutting edge equipment. Within the goal, we had two main aims. In Aim one we proposed to use the equipment to enhance five current research programs, which included the study of the evolution of antibiotic resistance, understanding the biological and behavioral consequences of chronic sleep restriction on emotion processing, developing novel bioinsecticides against mosquitoes, understanding how neural toxins affect neurogenesis and promoting comparative genomics research through the Global Invertebrate Genomics Alliance (GIGA) and microbial metagenomics. In Aim two, we proposed to enhance the quality and capabilities of current courses and allow development of leading edge STEM curricula. In the summary below, we break down our accomplishments by each piece of equipment requested. Furthermore, we outline how they were used in research, teaching and outreach. Furthermore, in the Appendices, we show that this equipment has been used to prepare manuscripts (Appendix 1) and grant proposals (Appendix 2), which will continue to enhance our research, teaching and outreach capabilities. Overall, we believe that we have met or exceeded the goals outlined in our original project narrative.

Equipment requested by Principal Investigator Robert P Smith

VictorX4 Microplate Reader from Perkin Elmer (and filters)
A) Use of new equipment in research

The VictorX4 Microplate reader has continued to see heavy use. As evidence of this we have already had to replace the bulb in the instrument as it began to wear out. This instrument has been used primarily in three projects that are centered on antibiotic resistance and preventing the spread of infectious disease. These projects include 1) uncovering how the inoculum effect can drive spontaneous antibiotic resistance, 2) examining which parameters are critical in allowing the inoculum effect and 3) characterizing engineered bacteria that may be useful towards eliminating parasitic nematode populations. In addition to these projects, Dr. Joshua Loomis has used this instrument for ELISA assays using enzymes from marine organisms.

In research project 1, we have used the microplate reader to grow bacteria under conditions that lead to the inoculum effect (Figure 1). Next, we have removed the bacteria and examined each population for antibiotic resistance mutants. We continue to see interesting relationships between dosage and mutation frequency in the inoculum effect region. Repeated experiments (performed by a different student) have shown the biphasic trend presented in our interim report. Now, we are focusing on perturbing the inoculum effect region (by applying heat shock or by applying protease inhibitors). Thus far, we continue to see a biphasic trend of mutation frequency and increasing mutation frequency at increasing concentrations of kanamycin
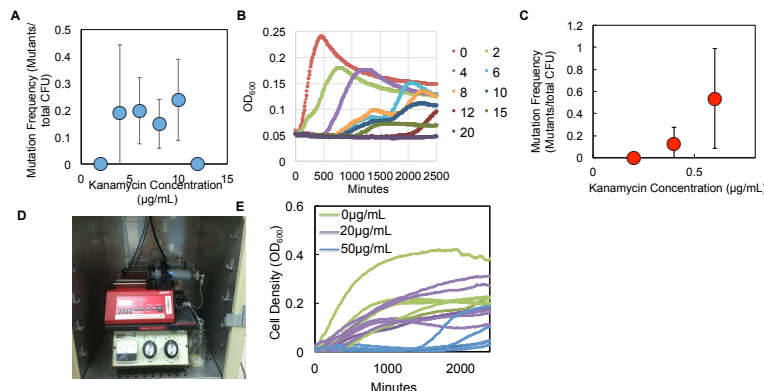

Figure 1: Representative data generated using the Victor X4 microplate reader. A) Mutation frequency of cell growing in the inoculum effect. B) Representative growth profiles of cells (OD_{600}) the inoculum effect region. C) Perturbing the inoculum effect region using heat shock produces mutants. D) Using an additional grant, we recreated a flow system to examine periodic applications of antibiotics and the inoculum effect. E) Growth profiles of mutants isolated after periodic application of kanamycin at a pulse period that leads to inoculum effect.

when the cells are heat shocked. However, the concentrations of kanamycin that this occurs at are significantly lower that when the cells are not heat shocked. This indicates that the antibiotic resistance mutants can evolve at lower kanamycin concentrations. We will continue to examine this in the coming months.

In addition to this project, we have also completely reconstructed a microfluidic flow system (funding from another grant). Using this flow system, we have pulsed the antibiotic kanamycin into a population of *E. coli*. As previously observed, this population of *E. coli* is able to grow at intermediate periods of antibiotic application, but not long or short applications. Next, we

isolated mutants from cultures grown in under intermediate pulse periods and quantified mutation frequency. Here, we observed that growth in this region led to antibiotic resistant bacteria, where resistance is not due to the inoculum effect, but is probably acquired through genetic mutation. We then isolated several mutants and grew the cells in the microplate reader at increasing concentration of kanamycin. We observed that a subset of these mutants were resistant to up to 50μg/ml of kanamycin, which represents a near 5-fold increase the kanamycin resistance (as compared to the wildtype strain).

In research project 2, we have continued to use the microplate reader to examine growth of our engineered bacteria that have the core behavior of the inoculum effect: population level bistability (Figure 2A and B). Here, at a sufficiently low density of bacteria, the population dies. Otherwise, if the density is sufficiently high, the population lives. Building on our results presented in the interim report, we have been able to elucidate principles that govern survival and growth as the spatial structure of the population is disturbed. By growing our engineered bacteria in the microplate reader, we have been able to uncover unique principles that may allow the identification of unique treatment options of bacterial infections. Furthermore, our results may have implications in controlling the spread of infectious diseases and the spread of invasive organisms.
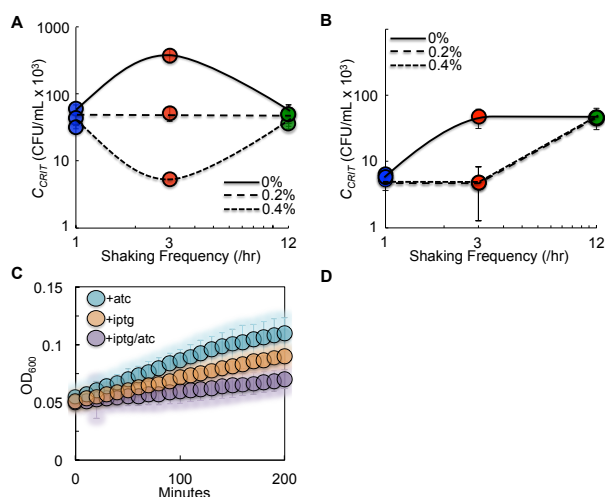

Figure 2: Using the Victor X4 microplate reader to examine population level bistability using a synthetic strain of bacteria. A) Shaking the plate at various frequencies alters the density required for our engineered bacteria to grow. B) Stabilizing cooperation amongst our engineered bacteria reduces the effect of shaking on the density of bacteria required for growth. C) Examining growth rate of engineered bacteria (designed to killed nematodes) expressing various components of our synthetic gene circuit.

In research project 3, we aim to develop a strain of engineered bacteria that is able to destroy parasitic nematode populations (Figure 2C). The microplate reader has been critical in ensuring functionality of the engineered bacteria. Specifically, we have extended the use of the microplate reader to this project by quantifying the effect of synthetic gene circuits (that aim to kill nematodes) on bacterial physiology and to ensure circuit functionality.

B) Integration with teaching

The VictorX4 microplate reader has been used in BIOL 3400, Microbiology/Lab. Here, students grew different strains of bacteria and examined growth curves (sample image provided in last report). This instrument has also been integrated into BIOL 3600, Genetics/Lab, where students examined fluorescent proteins and engineered bacterial growth (Figure 3). Overall, approximately 120 students have used this equipment in teaching labs to date. Use in teaching labs will continue in the future.
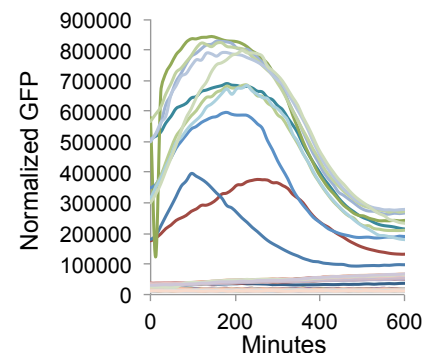

Figure 3: Representative GFP expression curves generated by students in a BIOL 3600 class.

Students in independent studies and Honor's thesis classes have also used the equipment.

C) Outreach with equipment
In the summer semester of 2014, one student from Miami Dade College used this equipment to quantify bacterial growth. In the coming summer semester, our outreach with this equipment will continue as we anticipate using this equipment once again with Miami Dade College students.

10L Incushaker from Genesee Scientific
A) Use of new equipment in research
This instrument has been used extensively to growth cells for all three projects described above. Furthermore, the refrigeration component has been used to culture cells for DNA transformation purposes. Similar to the microplate reader, this instrument has been used on a daily basis since being acquired in August.

B) Use of new equipment in teaching
We have also used this incubator to grow cells for microbiology (BIOL 3400), introductory biology (BIOL 1500) and genetics (BIOL 3600) classes. Students in independent studies and Honor's thesis classes have also used the equipment.

Renovation to the Sunken Building

A-C) Renovations to the Sunken Building were completed the week of August 11th, 2014. This room now houses the fluorescent microscope and the microplate reader. We have made extensive use of the equipment housed in this room for both research and teaching exercises. Outreach with this renovated room will likely be accomplished this summer in conjunction with a Miami Dade College program.

ChemiDocXRS+ Gel Imaging System from BioRad

A) Use of new equipment in research
This instrument was received at the end of August and was placed in a centrally located area so that multiple researchers and classes can make use of the gel imaging and chemiluminescent detection components of this system. We have made use of this instrument by visualizing PCR and restriction digests for several projects listed in this report. The sensitivity of this instrument has increased our capability to find very faint DNA bands, which has been critical in several projects. Furthermore, we have used this instrument to image one western blot that was stained with a chemiluminscent antibody.

Dr. Aarti Raja has also used this equipment for her research into antibiotics (Small World's project described below) and Dr. Emily Schmitt has used it to visualize cDNA as she investigates how common additives, such as aspartame, affect gene expression in yeast cells. Without this equipment, visualization of weak DNA signals would be quite challenging (especially for the Small World's project).

B) Use of new equipment in teaching

This gel imaging system has received extensive use in teaching. Specifically several introductory biology (BIOL 1500) and genetics (BIOL 3600) classes have used this instrumentation to visualize a wide range of agarose gels (PCR, RFLP, cloning etc., Figure 4). We estimate that over 400 students have made use of this equipment to date. This instrument will continue to be used heavily in the years to come. Furthermore, we will likely extend its use to biochemistry class as we are currently piloting the visualization of chemiluminescent antibodies in the instrument. Students in independent studies and Honor's thesis classes have also used the equipment.
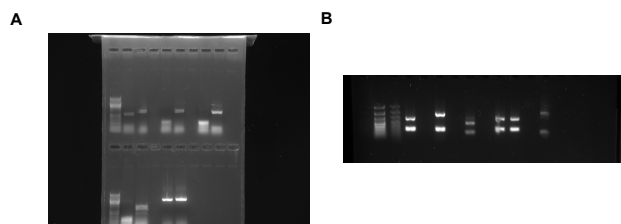

Figure 4: Gel electrophoresis images produced by students in BIOL 3600 (Genetics) and BIOL 1500 (Introductory Biology). A) Students used the ChemidocXRS+ to visualize RFLP gels. Representative gel image shown. B) Students used the system to examine PCR products generated in a crime scene simulation lab.

C) Use of the new equipment in outreach

Dr. Aarti Raja and Dr. Chris Blanar have used this instrument in conjunction with the Small World's project hosted by Yale University. Here, students isolated bacteria from soils and screen them for antibacterial properties. Bacteria were identified using PCR, which were visualized using this instrument.

Equipment requested by Key Personnel Jose V. Lopez

A) Use of new equipment - Illumina MiSeq Results

At the time of writing, our molecular genetics laboratory at the NSU Oceanographic Center has performed more than seven successful MiSeq DNA sequencing runs of 16S rRNA amplicons (Fig 5-7). More than 16.32Gb of DNA sequence data has been generated from this MiSeq instrument. Data is automatically downloaded to Illumina's Basespace platform (https://basespace.illumina.com), where quality can be checked, and further analyses can be performed. Example output from one of our experiments is shown in Figure 6. The QScore panel in the upper right indicates that the data quality is excellent, with >93% of the reads showing a Phred score of Q30 or above.
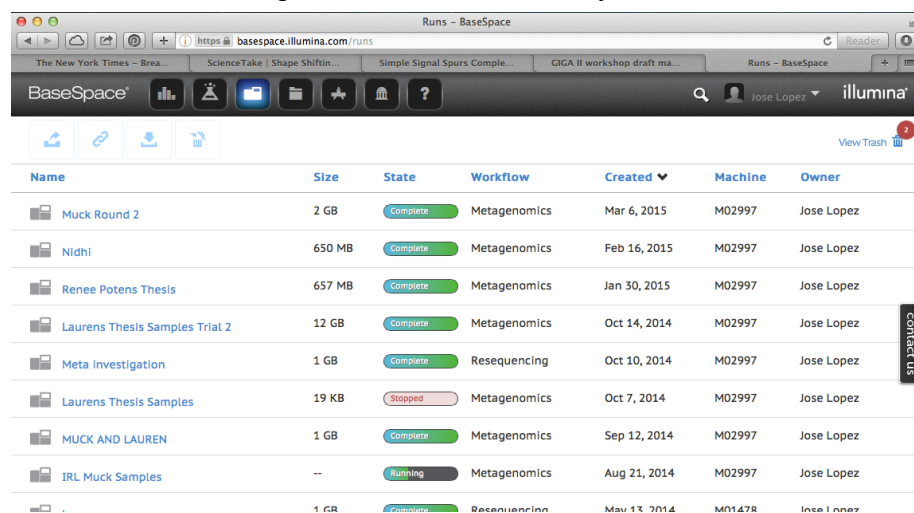
B) Integration with Teaching (and Mentoring)

These runs have mostly been performed by three NSU Masters level graduate students, who followed a modified 16S rRNA workflow that utilized universal bacterial primers (Caparaso et al, 20111, Figures 5-7). Each MS student has a unique thesis project in the Lopez laboratory that usually has aims to

GIGA.Nova.edu, Earthmicrobiome.org and www.Deependconsortium.org

Illumina MiSeq technology has entered our laboratory for high throughput DNA sequencing and training of students

Figure 5: The MiSeq instrument is a valuable tool for training graduate and undergraduate students the latest in "next generation" DNA sequencing methods.

characterize the alpha and beta diversity of microbial communities (also known as "microbiomes") from specific habitats, ranging from local marine waters to marine muck to coral reef sponge species to vertebrate skin or fecal samples on swabs. Depending on their habitat and the questions being addressed, typically only 1-2 MiSeq runs is needed to provide each student with sufficient sequence data to meet

Figure 6: A representative screenshot for one of our most recent experiments.

their thesis objectives. Following each run, students then also independently perform analyses and interpretation of the data, with the aim of publishing their results in peer-reviewed manuscripts. We have also been training one undergraduate FCAS student in the preparation of samples for the MiSeq, which will continue the rest of the year through a directed individual study (DIS) project with Professor Lopez.

We will likely mentor new undergraduate interns this summer and train them on techniques, which will be applicable to DNA sequencing methods and this instrument.

We also successfully integrated the operation of the MiSeq instrument and data analysis in the teaching of a Fall 2014 semester graduate level course at the Oceanographic Center - Genomics (OCMB 8020). This course discussed and exhibited the MiSeq workflow to all students.

C) Outreach with equipment

We have already begun projects and collaborations utilizing the MiSeq through various grant funded and local agencies such as Broward County Environmental Protection and Growth Management Department (EPGMD), the Broward County Sherrif's Office and DNA Crime lab, and the GOMRI (Gulf of Mexico Deep Pelagic Nekton Dynamics) - http://www.deependconsortium.org, ORCA (Ocean Reconnaissance and
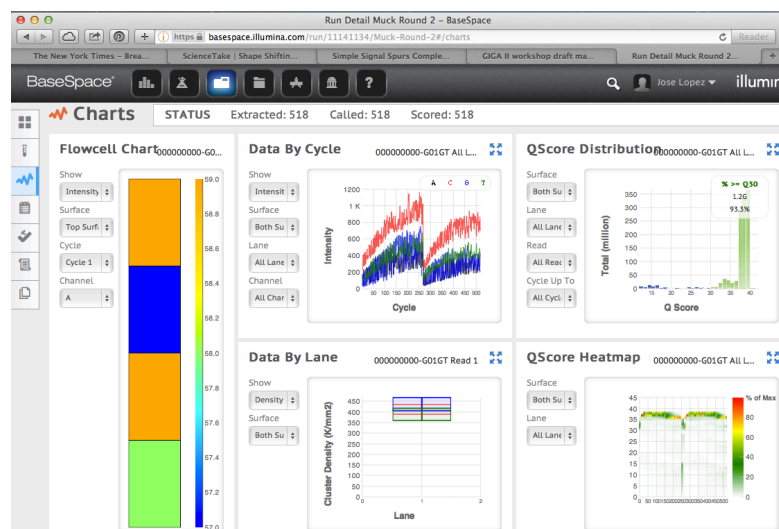

Figure 7: Representative image showing high quality DNA and sequencing data obtained from the MiSeq.

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Conservation Association - www.team**orca**.org/), Florida Atlantic University and the Global Invertebrate Genome Alliance (GIGA.Nova.edu).

Complete Computational Cluster
A-C) Use of new equipment in research, teaching and outreach
The computational cluster has been installed and now being used by the Lopez laboratory. There is over 14 Tb of storage space, which will mostly hold MiSeq output and data analyses generated by downstream programs. This cluster will greatly enhance our ability to assemble genomic sequences and will be used in conjunction with the goals of the MiSeq Sequencer.

CFX96 qPCR machine from BioRad
A-C) Use of new equipment in research, teaching and outreach
The CFX96 qPCR machine has been used several times, though a student has not formally incorporated it into a specific project.

Qubit from Life Sciences
A-C) Use of new equipment in research, teaching and outreach
We received the Qubit and have used it to quantify DNA. This instrument will has been used along with the MiSeq.

Freezer from Fisher Scientific
A-C) Use of new equipment in research, teaching and outreach
This freezer was recently installed and is currently being used to house some of our DNA samples. In the future, it will be used to house additional samples for the MiSeq and related activities.

Equipment requested by Key Personnel Jaime Tartar

Qiagen QIAcube
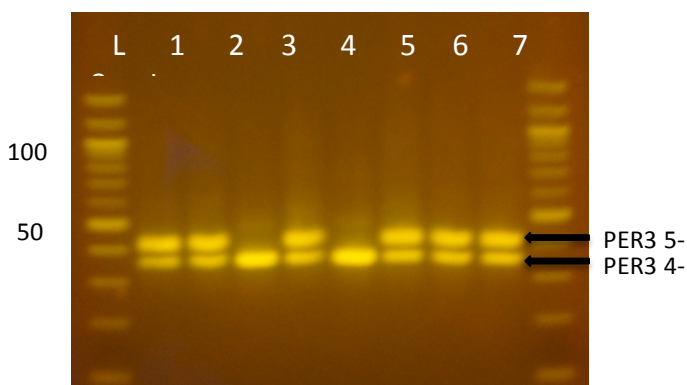
Figure 8: Data showing PCR products from students in Introduction to Neuroscience Lab. Student DNA was extracted with the QIAcube. Agarose gel shows VNTR polymorphisms in the PER3 gene. This lab teaches students about Clock genes and the molecular basis of individual differences in circadian rhythms.

A) Use of new equipment
The Qiagen QIAcube has been fully integrated into various teaching and research activities. We have extracted DNA from over 200 participants in three studies aimed at investigating how genetic polymorphisms contribute to individual differences in behavioral health after sleep loss. In addition, this equipment has been used by Key Personnel Aurelien Tartar to perform DNA purification for multiple PCR protocols.

B) Integration with teaching

The QIAcube has been integrated into NEUR 2500 "Introduction to Neuroscience" and NEUR3000, "behavioral genetics" classes. In both classes, students extract their own DNA in and then carry out various PCR protocols. So far we have used the QIAcube for this purpose in two Introduction to Neuroscience classes and one Behavioral Genetics class (Figure 8). Moreover, the use of this equipment has spurred the development of a proposal to increase genetics research in these classes. With the ease of DNA extraction and purification using the QIAcube we have proposed to develop a student discovery-based research program at NSU- the NSU Genetic Institute for Undergraduate Students (NSU GenIUS). Fundraising for this program is underway and the program will be offered as a discovery-based lab course for students. Finally, the QIAcube was also used to extract DNA in a BIOL 3600 class.

C) Outreach with equipment

Key Personnel Tartar has recently received a funding award from the Department of Education (Number P120A140012) where she serves as the Co-PI on a project aimed at increasing student research at a local 4 year college (Miami Dade College, MDC). As part of this SPARC Science Peer and Research Collaborative (SPARC), several students from MDC will work on the NSU campus over the next three summers and will extensively use the QIAcube as part of a series of research projects aimed at identifying biomarkers and polymorphisms associated with normal and impaired sleep behavior. The ability to plan and carry out these studies was greatly aided by the acquisition of the QIAcube.

Invitrogen Qubit

A) Use of new equipment

The Qubit has been received and installed into the psychophysiology laboratory. Although this equipment has not been applied to research or classroom purposes yet, it will be used this summer as part of Key Personnel Tartar's summer training program with MDC students. This equipment also supports the GenIUS program development.

Phillips Actiwatches

A) Use of new equipment

The Actiwatches have been fully integrated into several ongoing research projects. These projects involve investigating chronic sleep restriction on various types of health measures. The actigraphy allows us to non-invasively, but objectively, record sleep behavior. These measures are related back to several biomarker and neurobehavioral measures of health and performance. Initial findings from the use of these watches was used by Key Personnel Tartar as part of pilot data for a recent NIH Academic Research Enhancement Award (AREA) Program (R15) application that was submitted in February 2015 (1 R15 CA202626-01, An Integrative Analysis of Chronic Sleep Restriction's Effect on Cancer Risk).

B) Integration with teaching

As part of a credit-bearing research class (Independent study in Neuroscience), several students have already been trained on how to use the actiwatches and analyze actigraphy data for the study on sleep restriction.

C) Outreach with equipment

The above research projects are conducted in collaboration with two NSU centers: the NSU Center for Psychological Studies and the Neuro-Immune Institute. In addition, these watches are used as part of Tartar's Department of Education grant (see QIAcube outreach) where college students from Miami Dade College will receive training on the actiwatches as part of their summer research training program.

Compumedics EEG

A) Use of new equipment

Faculty members engage both graduate and undergraduate students in research. Students in the Electroencephalography (EEG) Laboratory learn the principles of EEG recording and investigate the effects of sleep deprivation and emotional processing on cognitive tasks. Yet other students investigate inhibitory control in monolingual and bilingual speakers. Students present their findings in our Undergraduate Student Symposium, which is held on campus every year. Moreover, their work is presented at national conferences and often leads to publications in peer-reviewed journals.

B) Integration with teaching

The multichannel EEG is used to actively engage students in the learning process and to teach students about brain-behavior relationships. Students use this state-of-the-art equipment to learn principles of electroencephalography in Psychophysiology (PSYC 4400). They work in groups to design research studies, to record the brain's electrical activity, and to learn the link between electroencephalography and behavior. Moreover, students prepare oral presentations to share their results with their classmates. Together, the experience of designing research projects, collecting data, and presenting their findings helps students to develop critical thinking and research skills. This equipment is also used for class demonstration of EEG in Research Methods in Neuroscience (NEUR 2700). During this lab activity, students are able to records their own EEG equipment and learn how distinct EEG bands correspond to different state of arousal.

C) Outreach with equipment

Every year NSU holds Brain Awareness Week, a weeklong event in which students and faculty give lectures and live demonstrations to inform the community of the latest research in brain sciences. This event is widely advertised and people of all ages from the NSU community, the Lifelong-Learning Institute and from the community at large are invited to the main campus. This year, students used our newly acquired EEG equipment to demonstrate the brain's electrical activity and to discuss changes in the brain as a function of aging and illness.

Equipment requested by Key Personnel Aurelien Tartar

Lightcycler 96 from Roche
A) Use of new equipment in research

The Light Cycler 96 was received from Roche on June 19, 2014. This date coincided with the beginning of A. Tartar's sabbatical leave and therefore the installation and training by the Roche technicians were delayed until January 2015 (end of Tartar's sabbatical). The technicians were very understanding, and initiated the machine set-up in January 2015. The machine did not require any visit from the ROCHE Field Application Specialist (Charles Hardwick, Ph.D.) but was installed and operational after several brief email exchanges and phone conversations between Hardwick and Tartar. The associated supplies, such as qPCR 8-tube Strips and FastStart Essential DNA Green Master kit, were
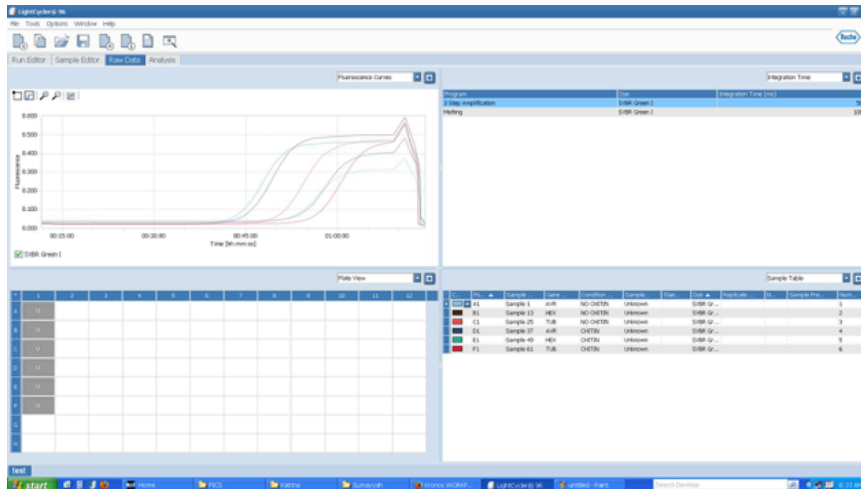

Figure 9: A screenshot of the installed LightCycler96 Application software demonstrating that both the machine and the software have been tested and are now routinely used in the Tartar laboratory.

subsequently ordered from Roche and received in February 2015. The machine was tested and no issue was detected, allowing for Tartar and selected undergraduate students to become familiar with both the machine and the analysis software (Figure 9).

Our plan remains to train the undergraduate research assistants on the logical continuation of the research project that was detailed in the grant proposal. Most of the data generated by previous undergraduate research assistants has now been published (Quiroz Velasquez et al., 2014), and consists
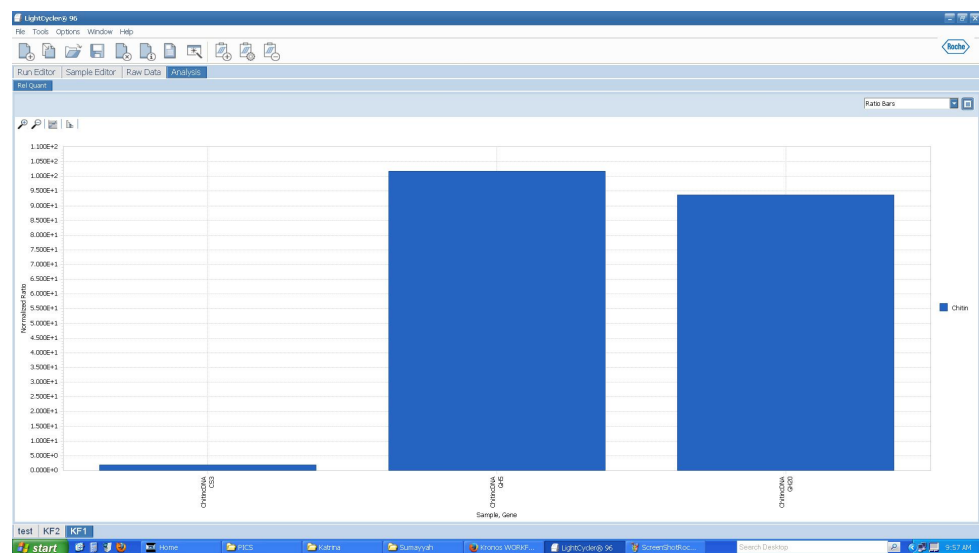

Figure 10: Preliminary differential expression analyses performed using the ROCHE Lightcycler 96 indicated that the *L. giganteum* GH5_27 and GH20 genes are overexpressed in the presence of chitin, suggesting a biological activity against mosquitoes.

of several promising proteins identified in the *Lagenidium giganteum* transcriptome. These predicted proteins may have insecticidal activities on mosquito larvae and therefore may be developed as novel bioinsecticides against mosquitoes. Currently, we are focusing on two selected *L. giganteum* proteins, corresponding to Glycoside Hydrolases family 5 and family 20 (GH5 and GH20, respectively). The GH5_27 enzyme (GH5 subfamily 27) was described in our recent publication (Quiroz Velasquez et al., 2014), while the characterization of the GH20 enzyme remains unpublished (a manuscript is in preparation and is scheduled to be submitted for publication during June/July 2015). Our objective is to use the real time thermocycler in order to determine the gene expression pattern of these candidate genes during the infection process, and initiate the functional characterization of the GH5_27 and GH20 proteins. Toward this end, the oomycete *Lagenidium giganteum* was grown in two distinct conditions. One condition consisted of a traditional artificial media (Peptone + Yeast Extract + Glucose, or PYG) as previously described (Kerwin and Petersen, 1997), whereas the second condition consisted of PYG media supplemented with chitin (commercially available from Sigma) at a concentration of 1g per 50 mL of media. After 4 days of growth, RNA was extracted from both sample conditions and cDNA was generated. The cDNA samples were used as templates for qPCR reactions that included GH5_27 and GH20 gene specific primers, as well as appropriate controls (tubulin and cellulose synthase gene). As indicated in Figure 10, preliminary data obtained using the LightCycler 96 indicated that both GH5_27 and GH20 genes are differentially expressed in the presence of chitin, suggesting that these proteins may play a role in the pathogenicity process and have biological activities against mosquitoes.

The data presented in Figure 10 is currently being replicated and confirmed. This work is performed by the undergraduate students working in Tartar's laboratory. The current team of students consists exclusively of minority students. A publication is anticipated for late 2015.

Our plan is to continue our gene expression analyses in the summer 2015, while expanding our current activities. More specifically, the ROCHE Lightcycler 96 will be used to performed SNP genotyping assays in an effort to assist coPI Jaime Tartar's project. The Taqman-based assay from Life Technologies will be purchased and used for this project.

B) Integration with teaching
Integration of the equipment with teaching activities is scheduled for Fall 2015. Following extensive training and use of the RT-PCR system in the Winter and Summer 2015 for our research program(s), the equipment will be introduced in two laboratory based undergraduate classes: BIOL 3600 (Genetics) and BIOL 4100 (Genomics).

C) Outreach with equipment
Outreach/volunteers have been planned for Summer 2015. Specifically, Tartar was the recipient of Department of Education award (Minority Science and Engineering Improvement Program/MSEIP), and will host minority undergraduate students from Miami Dade College in Summer 2015, 2016 and 2017. These students will be introduced to the thermocycler and have the opportunity to develop research projects, or contribute to on-going research projects, that used this machine.

Equipment requested by Key Personnel James Munoz

Olympus Fluorescent Microscope
A) Use of new equipment
The Olympus fluorescent microscope has been fully integrated into research and teaching activities. Prior to obtaining this microscope, we had to schedule time at the medical school facility and transport our samples between facilities. Having access to this microscope on our main campus has increased our research productivity by increasing the ease of access for our students and the amount of time we can spend on the microscope. The microscope has been used to generate data examining the effects of agonism of angiotensin II AT1 receptor and AT2 receptors on neural proliferation, differentiation, and survival in human-derived neural stem cells. These data have recently been presented at regional and national meetings including the SYNAPSE meeting at the Max Planck Institute and the American Junior Academy of Science and AAAS meeting. We anticipate publication of the data by mid-summer and will properly acknowledge the DoD funding. PI Smith has also used this microscope to image cells in all three project described in his section. He has made heavy use of the fluorescent capabilities of this microscope by examining reporter genes/assays. He has also used it to image and quantify the movement of nematodes

Figure 11: Representative screenshot of measurements of C. elegans movement using the fluorescent microscope.

(Figure 11). Results from this study were published in conference proceedings with an undergraduate student first author.

B) Integration with teaching
The fluorescent microscope has been integrated into NEUR 2700 "Methods in Neuroscience." Students in this class prepared and processed brain sections from spontaneously hypertensive rat brains and wild-type, age-matched controls to pursue student-derived hypotheses. Preliminary data generated by some of these projects will be pursued through independent study or honors theses with these students (Figure 12).
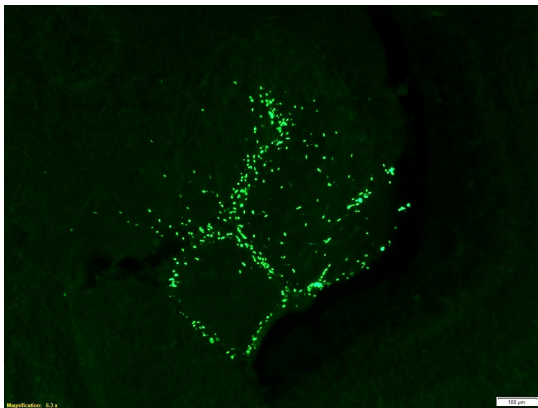

Figure 12: Representative image taken by students using the fluorescent microscope.

Centrifuge
A) Use of new equipment
The high speed eppenddorf 5804R centrifuge has been integrated into research projects. Most recently, the centrifuge has primarily been used to isolate RNA and protein. RNA was isolated from human-derived neural stem cells for the purpose of conducting real-time RT-PCR to examine changes in gene expression following agonism of the angiotensin II AT1 and AT2 receptors. Findings from this study will be used to further our investigation into how agonism of these receptors promotes either cell survival or cell death. This equipment will become integral to our research

projects aimed at examining how toxins affect gene and protein expression in maturing brain cells.

B) Integration with teaching
The centrifuge has been integrated into NEUR 2700 "Methods in Neuroscience." Students in this class isolated protein from spontaneously hypertensive rat brains and wild-type, age-matched controls to pursue student-derived hypotheses. Preliminary data generated by some of these projects will be pursued through independent study or honors theses with these students. Finally, this centrifuge was used in the fourth year parasitology course to isolate nematode eggs from a complex soil mixture.

Hood and Incubators
A) Use of new equipment
The SterilGARD® 504 e^3 and Galaxy CO-170S cell culture incubators is the one piece of equipment providing us with the greatest independence and advancing several research projects. The previous lack of cell culture facilities prevented several faculty from being able to pursue or advance their research programs. Several faculty have expressed an interest in using the equipment. For instance, a new faculty member starting this fall will be bringing dental pulp stem cells, collaborations will continue with pharmacology faculty to examine effects of agonism of the angiotensin II AT1 and AT2 receptors on neural stem cell development, and experimental plans are being developed to initiate studies examining how toxins affect maturing brain cells. Note that the funding requested to upgrade electrical capability has ensured functionality of this equipment.

B) Integration with teaching
The cell culture hood and incubators has been integrated into NEUR 2700 "Methods in Neuroscience." Current students learned basic principles of cell culture. During the next course offering, students will be culturing neural stem cells and pursing student-derived hypotheses that may be further developed through independent study or an honors thesis. This equipment is also being used to prepare students for summer internships, in which cell culture may be used. For instance, one of our students has been selected for a summer internship at Harvard Medical School where he will be examining HIV entry into cells. We are preparing him for the internship by teaching him basic cell culture techniques.

Outreach with equipment (same for all equipment requested by Key Personnel Munoz)

Nova Southeastern University has partnered with Life Sciences of South Florida with the goal of creating a South Florida life science consortium to foster collaboration, enhance educational opportunities, and increase access to and utilization of expensive instruments and computers. Current potential institutions that may be involved in the outreach with this microscope include 15 public and private colleges and universities in South Florida. Additionally, Key Personnel Munoz has participated in the U.S. Department of Education STEM Ladder Grant aimed at increasing student research at Miami Dade College (MDC), a local 4-year college. As part of this initiative, students from MDC will work alongside our independent study students over the summer to develop the proposals put forth in the grant.

Appendix 1: Manuscripts and theses currently in preparation that have used the equipment acquired in this grant.

Bracho, O.R., Manchery, C., Haskell, E., Blanar, C.A., and Smith, R.P. A synthetic nematode biocontrol agent reveals hidden implementation constraints. Pending submission to *Molecular Systems Biology*.

Chayo, I, Sandor, S. Fins, A.I., Tartar J.L. The Late Positive Potential ERP response to an emotion task is sensitive to the accumulation of homeostatic sleep pressure. In preparation

Fins, K.C., Abiff, S.K., Tartar, A. A *Lagenidium giganteum* Glycoside Hydrolase family 20 (GH20) is a novel virulence factor against mosquito larvae. In preparation

Hill, L. The Effect of implicit Emotion on Electrophysiological Measures of Emotion Processing. MS Thesis in Preparation

McKissack, H., Mohammed, U., Nemzer, L., and Smith, R.P. The inoculum effect provides opportunistic conditions for the evolution of antibiotic resistant bacteria. In preparation.

O'Connell, L. Compositional Analysis of the Port Everglades Inlet (Broward County, FL) Microbiome Using Next Generation Sequencing Technology, MS Thesis in preparation.

Potens, R. Genetic Analysis of Seasonal Fluctuations in Microbial Symbiont Diversity of the South Florida Marine Sponge *Amphimedon compressa*, MS Thesis in preparation.

Viena, T., Gobin, C.M., Fins, A.I. Craddock, T.J. Tartar A., Tartar J.L. Per3 4/4 polymorphism combined with short sleep duration is associated with increased anxiety and depressive symptomatology. In preparation.

Vijayan, N. Analysis of bacterial diversity with response to light and antibiotics in in vitro tissue culture of marine sponge, Cinachyrella, MS Thesis in preparation.

Wilson, C.E. Examining quorum sensing and diffusion sensing using engineered bacteria. MS Thesis in preparation

Wilson, C.E., Driscoll, W., Eldakar, O.T., Lopez, J., and Smith, R.P. Spatial disturbance as a mechanism to unite quorum sensing and diffusion sensing. Pending submission to *Nature*.

Appendix 2: Grant proposals submitted/in preparation using preliminary data acquired using the equipment

Smith, RP. Engineering a Trojan horse: using synthetic biology to prevent nematode infections. National Institutes of Health Academic Research Enhancement Award (AREA) Program (R15) (submitted June 2014, invited resubmission in June 2015).

Smith, RP. Using engineered bacteria to prevent microbial cooperation. Nova Southeastern University Internal Research Grant (submitted March 2015).

Smith, RP. Using synthetic biology to create nematode killing engineered bacteria. United States Department of Agriculture AFRI/NIFA Program (submitted April 2015).

Tartar, A. Co-PI: Robert P Smith. A genomic and synthetic biology approach to identify and assess novel biopesticides. United States Department of Agriculture AFRI/NIFA Program (submitted April 2015).

Tartar, J. (Co-PI) An Integrative Analysis of Chronic Sleep Restriction's Effect on Cancer Risk (GRANT11846390) National Institutes of Health Academic Research Enhancement Award (AREA) Program (R15) (submitted February 2015).

Tartar, A, Co-PI: Robert P Smith. Developing second generation Lagenidium-based larvicides. Bill and Melinda Gate Foundation Global Challenges (to be submitted in May 2015).

REPORT OF INVENTIONS AND SUBCONTRACTS
(Pursuant to "Patent Rights" Contract Clause) (See Instructions on back)

The public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing the burden, to the Department of Defense, Executive Services Directorate (9000-0095). Respondents should be aware that notwithstanding any other provision of law, no person shall be subject to any penalty for failing to comply with a collection of information if it does not display a currently valid OMB control number.

PLEASE DO NOT RETURN YOUR COMPLETED FORM TO THE ABOVE ORGANIZATION. RETURN COMPLETED FORM TO THE CONTRACTING OFFICER.

	3. TYPE OF REPORT (X one)
	a. INTERIM b. FINAL [X]

1.a. NAME OF CONTRACTOR/SUBCONTRACTOR
Nova Southeastern University

b. ADDRESS (Include ZIP Code)
3301 College Avenue
Fort Lauderdale, FL 33314

c. CONTRACT NUMBER
W911NF-14-1-0070

d. AWARD DATE (YYYYMMDD)
20140201

2.a. NAME OF GOVERNMENT PRIME CONTRACTOR

b. ADDRESS (Include ZIP Code)

c. CONTRACT NUMBER

d. AWARD DATE (YYYYMMDD)

4. REPORTING PERIOD (YYYYMMDD)
a. FROM 20140201
b. TO 20150131

SECTION I - SUBJECT INVENTIONS

5. "SUBJECT INVENTIONS" REQUIRED TO BE REPORTED BY CONTRACTOR/SUBCONTRACTOR (If "None," so state)

NAME(S) OF INVENTOR(S) (Last, First, Middle Initial) a.	TITLE OF INVENTION(S) b.	DISCLOSURE NUMBER, PATENT APPLICATION SERIAL NUMBER OR PATENT NUMBER c.	ELECTION TO FILE PATENT APPLICATIONS (X) d.				CONFIRMATORY INSTRUMENT OR ASSIGNMENT FORWARDED TO CONTRACTING OFFICER (X) e.	
			(1) UNITED STATES		(2) FOREIGN		(a) YES	(b) NO
			(a) YES	(b) NO	(a) YES	(b) NO		
N/A	N/A							

f. EMPLOYER OF INVENTOR(S) NOT EMPLOYED BY CONTRACTOR/SUBCONTRACTOR

(1) (a) NAME OF INVENTOR (Last, First, Middle Initial)	(2) (a) NAME OF INVENTOR (Last, First, Middle Initial)
(b) NAME OF EMPLOYER	(b) NAME OF EMPLOYER
(c) ADDRESS OF EMPLOYER (Include ZIP Code)	(c) ADDRESS OF EMPLOYER (Include ZIP Code)

g. ELECTED FOREIGN COUNTRIES IN WHICH A PATENT APPLICATION WILL BE FILED

(1) TITLE OF INVENTION	(2) FOREIGN COUNTRIES OF PATENT APPLICATION

SECTION II - SUBCONTRACTS (Containing a "Patent Rights" clause)

6. SUBCONTRACTS AWARDED BY CONTRACTOR/SUBCONTRACTOR (If "None," so state)

NAME OF SUBCONTRACTOR(S) a.	ADDRESS (Include ZIP Code) b.	SUBCONTRACT NUMBER(S) c.	FAR "PATENT RIGHTS" d.		DESCRIPTION OF WORK TO BE PERFORMED UNDER SUBCONTRACT(S) e.	SUBCONTRACT DATES (YYYYMMDD) f.	
			(1) CLAUSE NUMBER	(2) DATE (YYYYMM)		(1) AWARD	(2) ESTIMATED COMPLETION

SECTION III - CERTIFICATION

7. CERTIFICATION OF REPORT BY CONTRACTOR/SUBCONTRACTOR (Not required if: (X as appropriate))   SMALL BUSINESS or   NONPROFIT ORGANIZATION

I certify that the reporting party has procedures for prompt identification and timely disclosure of "Subject Inventions," that such procedures have been followed and that all "Subject Inventions" have been reported.

a. NAME OF AUTHORIZED CONTRACTOR/SUBCONTRACTOR OFFICIAL (Last, First, Middle Initial)
Harlan, Catherine, M.

b. TITLE
Director, Office of Sponsored Programs

c. SIGNATURE

d. DATE SIGNED
4-28-15

DD FORM 882, JUL 2005   PREVIOUS EDITION IS OBSOLETE.